

CLAIMS

WHAT IS CLAIMED IS:

- 1) A method comprising:
 - a) obtaining one or more samples;
 - b) obtaining one or more types of target cells and/or microorganisms from each sample;
 - c) performing a bioluminescence regenerative cycle (BRC) assay to detect ATP and inorganic phosphate (PPi) in the cells and/or microorganisms; and
 - d) estimating the number of target cells and/or microorganisms from the BRC assay.
- 2) The method of claim 1, further comprising using one or more binding moieties to separate one or more types of target cells and/or microorganisms from the sample.
- 3) The method of claim 2, wherein the capture moiety is an antibody.
- 4) The method of claim 3, wherein the antibody is attached to a magnetic bead, glass bead, plastic bead, nitrocellulose membrane, nylon membrane, chromatography support or other solid surface.
- 5) The method of claim 1, further comprising lysing the cells and/or microorganisms.
- 6) The method of claim 5, further comprising partially purifying the ATP and PPi from the lysed cells and/or microorganisms.
- 7) The method of claim 1, wherein the sample is blood, serum, plasma, cerebrospinal fluid, lymphatic fluid, urine, stool, semen, lacrimal fluid, saliva, sputum, a biopsy sample, a tissue scraping, a swab sample, an endoscopic sample, a cell sample, a tissue sample, a food sample, a water sample, an environmental sample or an air sample.
- 8) The method of claim 1, further comprising accumulating the photon count over a time interval.
- 9) The method of claim 1, further comprising adding adenylate kinase and AMP substrate or pyruvate kinase and phosphoenolpyruvate to the BRC assay.

- 10) The method of claim 1, further comprising adding between 0.01 and 10 attomole of purified pyrophosphate or ATP to each assay to reduce background light emission.
- 11) A kit for performing BRC assays comprising:
 - a) one or more BRC reagents; and
 - b) a standard solution.
- 12) The kit of claim 11, wherein the standard solution comprises a known amount of ATP and/or PPi.
- 13) The kit of claim 11, wherein the BRC reagents comprise at least one reagent selected from the group consisting of luciferin, luciferase, ATP sulfurylase, adenosine 5'-phosphosulphate and BRC buffer.
- 14) An apparatus comprising:
 - a) a light tight slide;
 - b) a plurality of chambers in the slide, the chambers connected by a mono-directional flow channel;
 - c) an inlet and an outlet connected to the flow channel; and
 - d) a sealed secondary inlet connected to each chamber.
- 15) The apparatus of claim 14, further comprising an affinity matrix in each chamber.
- 16) The apparatus of claim 15, wherein each affinity matrix is attached to at least one binding moiety.
- 17) The apparatus of claim 16, wherein the binding moieties are antibodies or capture oligonucleotides.
- 18) The apparatus of claim 17, wherein each chamber comprises at least one antibody and at least one capture oligonucleotide, both specific for the same pathogen.